

# Human STAT1 knockout HeLa cell lysate ab263837

## 5 图像

### 概述

产品名称	人STAT1 knockout HeLa cell裂解物
产品概述	Knockout cell lysate achieved by CRISPR/Cas9.
Parental Cell Line	HeLa
Organism	Human
Mutation description	Knockout achieved by using CRISPR/Cas9, 14 bp deletion in exon4 and 1 bp deletion in exon4 and 5 bp deletion in exon4.
Passage number	<20
Knockout validation	Sanger Sequencing, Western Blot (WB)
Reconstitution notes	To use as WB control, resuspend the lyophilizate in 50 µL of LDS* Sample Buffer to have a final concentration of 2 mg/ml. For reducing conditions, we recommend a final concentration of 0.1 M DTT. <i>*Usage of SDS sample buffer is not recommended with these lyophilized lysates.</i>

### 说明

**Lysate preparation:** Our lysates are made using RIPA buffer to which we add a protease inhibitor cocktail and phosphatase inhibitor cocktail (ratio: 300:100:10). *This means that the protein of interest is denatured.* If you require a native form of the protein please use the live cell version - found [here](#). Please refer to our lysis protocol for further details on how our lysates are prepared.

**User storage instructions:** Lyophilizate may be stored at 4°C. After reconstitution, store at -20°C for short-term storage or -80°C for long-term storage.

Access thousands of knockout cell lysates, generated from commonly used cancer cell lines. [See here for more information on knockout cell lysates.](#)

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### 经测试应用

适用于: WB

性能

存放说明 Store at -80°C. Please refer to protocols.

组件	1 kit
ab262786 - Human STAT1 knockout HeLa cell lysate	1 x 100µg
ab255929 - Human wild-type HeLa cell lysate	1 x 100µg

Cell type	epithelial
Disease	Adenocarcinoma
Gender	Female
STR Analysis	Amelogenin X D5S818: 11, 12 D13S317: 12, 13.3 D7S820: 8, 12 D16S539: 9, 10 vWA: 16, 18 TH01: 7 TPOX: 8,12 CSF1PO: 9, 10

靶标

**功能** Signal transducer and activator of transcription that mediates signaling by interferons (IFNs). Following type I IFN (IFN-alpha and IFN-beta) binding to cell surface receptors, Jak kinases (TYK2 and JAK1) are activated, leading to tyrosine phosphorylation of STAT1 and STAT2. The phosphorylated STATs dimerize, associate with ISGF3G/IRF-9 to form a complex termed ISGF3 transcription factor, that enters the nucleus. ISGF3 binds to the IFN stimulated response element (ISRE) to activate the transcription of interferon stimulated genes, which drive the cell in an antiviral state. In response to type II IFN (IFN-gamma), STAT1 is tyrosine- and serine-phosphorylated. It then forms a homodimer termed IFN-gamma-activated factor (GAF), migrates into the nucleus and binds to the IFN gamma activated sequence (GAS) to drive the expression of the target genes, inducing a cellular antiviral state.

**疾病相关** Note=STAT1 deficiency results in impaired immune response leading to severe mycobacterial and viral diseases. In the case of complete deficiency, patients can die of viral disease. Defects in STAT1 are a cause of mendelian susceptibility to mycobacterial disease (MSMD) [MIM:209950]; also known as familial disseminated atypical mycobacterial infection. This rare condition confers predisposition to illness caused by moderately virulent mycobacterial species, such as Bacillus Calmette-Guerin (BCG) vaccine and environmental non-tuberculous mycobacteria, and by the more virulent Mycobacterium tuberculosis. Other microorganisms rarely cause severe clinical disease in individuals with susceptibility to mycobacterial infections, with the exception of Salmonella which infects less than 50% of these individuals. The pathogenic mechanism underlying MSMD is the impairment of interferon-gamma mediated immunity whose severity determines the clinical outcome. Some patients die of overwhelming mycobacterial disease with lepromatous-like lesions in early childhood, whereas others develop, later in life, disseminated but curable infections with tuberculoid granulomas. MSMD is a genetically heterogeneous disease with autosomal recessive, autosomal dominant or X-linked inheritance.

**序列相似性** Belongs to the transcription factor STAT family.  
Contains 1 SH2 domain.

**翻译后修饰** Phosphorylated on tyrosine and serine residues in response to IFN-alpha, IFN-gamma, PDGF and EGF. Phosphorylation on Tyr-701 (lacking in beta form) by JAK promotes dimerization and subsequent translocation to the nucleus. Phosphorylation on Ser-727 by several kinases including MAPK14, ERK1/2 and CAMKII on IFN-gamma stimulation, regulates STAT1 transcriptional

activity. Phosphorylation on Ser-727 promotes sumoylation though increasing interaction with PIAS. Phosphorylation on Ser-727 by PKCdelta induces apoptosis in response to DNA-damaging agents.

Sumoylated by SUMO1, SUMO2 and SUMO3. Sumoylation is enhanced by IFN-gamma-induced phosphorylation on Ser-727, and by interaction with PIAS proteins. Enhances the transactivation activity.

ISGylated.

## 细胞定位

Cytoplasm. Nucleus. Translocated into the nucleus in response to IFN-gamma-induced tyrosine phosphorylation and dimerization.

## 应用

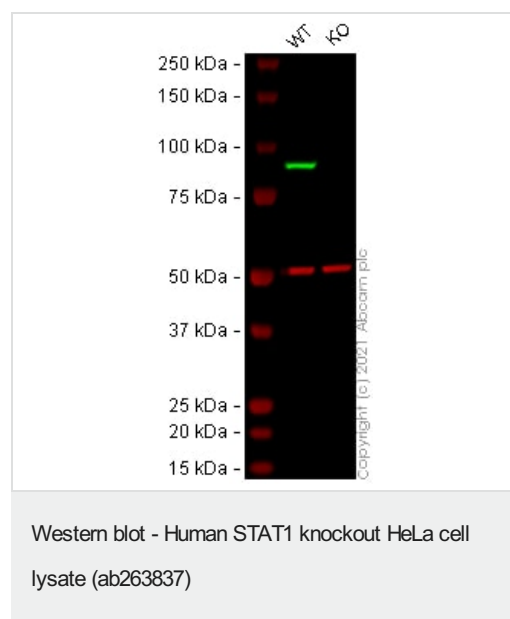
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“应用说明”部分 下显示的仅为推荐的起始稀释度;实际最佳的稀释度/浓度应由使用者检定。

应用	Ab评论	说明
WB		Use at an assay dependent concentration.

## 图片



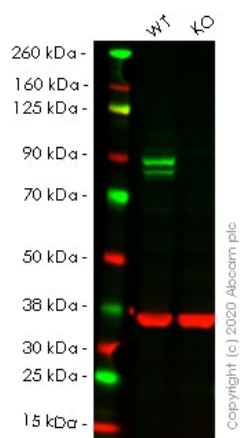
**Lane 1:** Wild-type HeLa cell lysate 20 µg

**Lane 2:** STAT1 knockout HeLa cell lysate 20 µg

**Lanes 1 - 2:** Merged signal (red and green). Green - **ab92506**

observed at 85 kDa. Red - loading control **ab7291** (Mouse anti-Alpha Tubulin [DM1A]) observed at 55kDa.

**ab92506** was shown to react with STAT1 alpha in wild-type HeLa cells in Western blot with loss of signal observed in STAT1 knockout cell line **ab255346** (STAT1 knockout cell lysate ab263837). Wild-type HeLa and STAT1 knockout cell lysates were subjected to SDS-PAGE. Membranes were blocked in 3 % milk in TBS-T (0.1 % Tween®) before incubation with **ab92506** and **ab7291** (Mouse anti-Alpha Tubulin [DM1A]) overnight at 4 °C at a 1 in 1000 dilution and a 1 in 20000 dilution respectively. Blots were incubated with Goat anti-Rabbit IgG H&L (IRDye® 800CW) preabsorbed (**ab216773**) and Goat anti-Mouse IgG H&L (IRDye® 680RD) preabsorbed (**ab216776**) secondary antibodies at 1 in 20000 dilution for 1 h at room temperature before imaging.



Western blot - Human STAT1 knockout HeLa cell lysate (ab263837)

**Lane 1:** Wild-type HeLa cell lysate (20µg)

**Lane 2:** STAT1 knockout HeLa cell lysate (20µg)

**Lanes 1- 2:** Merged signal (red and green). Green - **ab109320** observed at 87 kDa. Red - loading control **ab8245** observed at 37 kDa.

**ab109320** Recombinant Anti-STAT1 antibody [EPR4407] was shown to specifically react with STAT1 in wild-type HeLa cells in western blot. Loss of signal was observed when knockout cell line **ab255346** (knockout cell lysate ab263837) was used. Wild-type and STAT1 knockout samples were subjected to SDS-PAGE. Membrane was blocked for 1 hour at room temperature in 0.1% TBST with 3% non-fat dried milk. **ab109320** and Anti-GAPDH antibody [6C5] - Loading Control (**ab8245**) were incubated overnight at 4 °C at 1 in 10000 dilution and 1 in 20000 dilution respectively. Blots were developed with Goat anti-Rabbit IgG H&L (IRDye® 800CW) preadsorbed (**ab216773**) and Goat anti-Mouse IgG H&L (IRDye® 680RD) preadsorbed (**ab216776**) secondary antibodies at 1 in 20000 dilution for 1 hour at room temperature before imaging.

Mut	TGACAGGAGGT CATGAAAA - - - - - TGAAACATTTGGCAGCGTGCTCCCT
WT	TGACAGGAGGT CATGAAAAACGGATGGTGGCAAAATGAAACATTTGGCAGCGTGCTCCCT

Sanger Sequencing - Human STAT1 knockout HeLa cell lysate (ab263837)

Allele-1: 14 bp deletion in exon4

Mut	TGATCATCCAGCTGTGACAGGAGGT CAT - - - - - CGGATGGTGGCAAAATGAAACATCATTG
WT	TGATCATCCAGCTGTGACAGGAGGT CATGAAAAACGGATGGTGGCAAAATGAAACATCATTG

Sanger Sequencing - Human STAT1 knockout HeLa cell lysate (ab263837)

Allele-2: 5 bp deletion in exon4

Mut	TGTGACAGGAGT CATGAAAACG- ATGGTGGCAAATGAAACATCATTGGCAGCGTGCTCC
WT	TGTGACAGGAGT CATGAAAACGGATGGTGGCAAATGAAACATCATTGGCAGCGTGCTCC

Allele-3: 1 bp deletion in exon4

Sanger Sequencing - Human STAT1 knockout HeLa  
cell lysate (ab263837)

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